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U.S. Department of Agriculture Agricultural Research Service
August 1981

STA/STA

Agricultural Research

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Genetic Engineering— Limited Only by the Imagination

Genetic engineering, gene-splicing, recombinant DNA—whichever name is used—is becoming one of the most imposing tools acquired by science since the atom was split back in the days of World War II. Although cells have been swapping genes since life began, only recently have molecular biologists discovered ways to transplant specific genes to direct cells toward specific goals.

Hardly a week passes without a dramatic announcement of some new breakthrough in genetic engineering—interferon, an antiviral agent and a possible weapon against cancer; human insulin produced by bacteria to promise a steady supply for diabetics; human growth hormones to stimulate growth in stunted children; beta-endorphin, a natural pain killer found in the body; safe and inexpensive vaccines, such as the recently announced vaccine for foot-and-mouth disease, one of the world's most serious animal diseases (see *Agricultural Research*, September 1980).

Most of the dazzling advances in genetic engineering have been in the pharmaceutical area where technology is the most advanced. But within our

reach are more than vague promises of many other breakthroughs. The chemical industry will be making plastics, fuels, and chemicals from industrial wastes. Microorganisms will be designed to change inedible biomass into food and energy. The potential is enormous.

The agricultural sciences have also stepped across the threshold of the new technology of manipulating genes in the master molecule, DNA. Genetic manipulation, in the classical sense, is nothing new to agriculture. Plant and animal breeders have been manipulating genes for 50 to 60 years—a time- and space-consuming process that nevertheless paid off handsomely. The “green revolution” is an example of the results of that classical technology.

Just recently, ARS scientists in Beltsville, Md., using recombinant DNA techniques, found a way to identify a viroid in potato disease. From this, a commercial method can be developed to rid this crop of infected seed tubers. ARS and university scientists in Madison, Wis., have transplanted a gene, that directs protein production, from the French bean into a sunflower cell growing in a culture in the laboratory. The gene was transferred by means of the bacterium that causes crown gall disease, a plant cancer. This breakthrough is the first step toward a useful and versatile system for gene transfer between different plant species and genera.

But before we start planning “pork chops on trees,” a word of caution. The dramatic biomedical products catching the headlines today are relatively simple to produce, compared to transplanting genes or groups of genes between complex crop plants that may contain thousands of genes per plant. Producing interferon, human insulin, and pure vaccines requires transplanting only a single gene into a bacterium or yeast and then growing these

organisms in a laboratory vat where they produce the wanted product.

Tailoring crop plants to withstand drought, disease, high salt content of soil, and other adverse circumstances is a far more complex technology. It could involve more than a single gene to achieve a particular goal. Desirable traits are often the interaction of many genes. In addition to splicing a gene into the DNA of a bacterium, an additional gene transplanting step is necessary from the bacterium to a single cell of a crop plant. The plant cell with the new gene or genes must then be grown into a fully developed plant with leaves, roots, and all its parts to produce a crop. The technology for single-cell regeneration has not yet been mastered, but the work has begun. It is only a matter of time—not weeks or months, but years—before single-cell regeneration will become a reality for important crop plants.

The decades ahead will undoubtedly see more nutritious crops with their own disease resistance and resistance to environmental stress, crops that photosynthesize more efficiently, crops that fix their own nitrogen, and other innovative changes not yet on the drawing board. It is even possible that the shapes of crop plants that will feed the world 100 years from today will be different—perhaps so different that we today would find them hard to recognize.

The possibilities are limited only by the imagination!

By Virginia Dryden

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Agricultural Research
Vol. 30 No. 2
August 1981

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Agricultural Research is published monthly by the Agricultural Research Service (ARS), U.S. Department of Agriculture, Washington, D.C. 20250. The Secretary of Agriculture has determined that the publication of this periodical is necessary in the transaction of the public business required by law of this Department. Use of funds for printing this periodical has been approved by the Director of the Office of Management and Budget through June 15, 1982. Yearly subscription rate is \$13 in the United States and its territories, \$16.25 elsewhere. Single copies are \$1.25 domestic, \$1.60 foreign. Send subscription orders to Superintendent of Documents, Government Printing Office, Washington, D.C. 20402. Information in this magazine is public property and may be reprinted without permission. Prints of photos are available to mass media; please order by photo number.

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Magazine inquiries should be addressed to: The Editor, Information Staff, Room 3147-S, USDA, Washington, D.C. 20250. Telephone: (202) 447-6133.

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Cover: A gene from the seed of the French bean has been successfully transferred, via bacterium fragments, to sunflower tissue culture by ARS and University of Wisconsin scientists. John Kemp, ARS biochemist, inoculates a sunflower plant with *Agrobacterium* that is carrying the protein gene (068X597-15).

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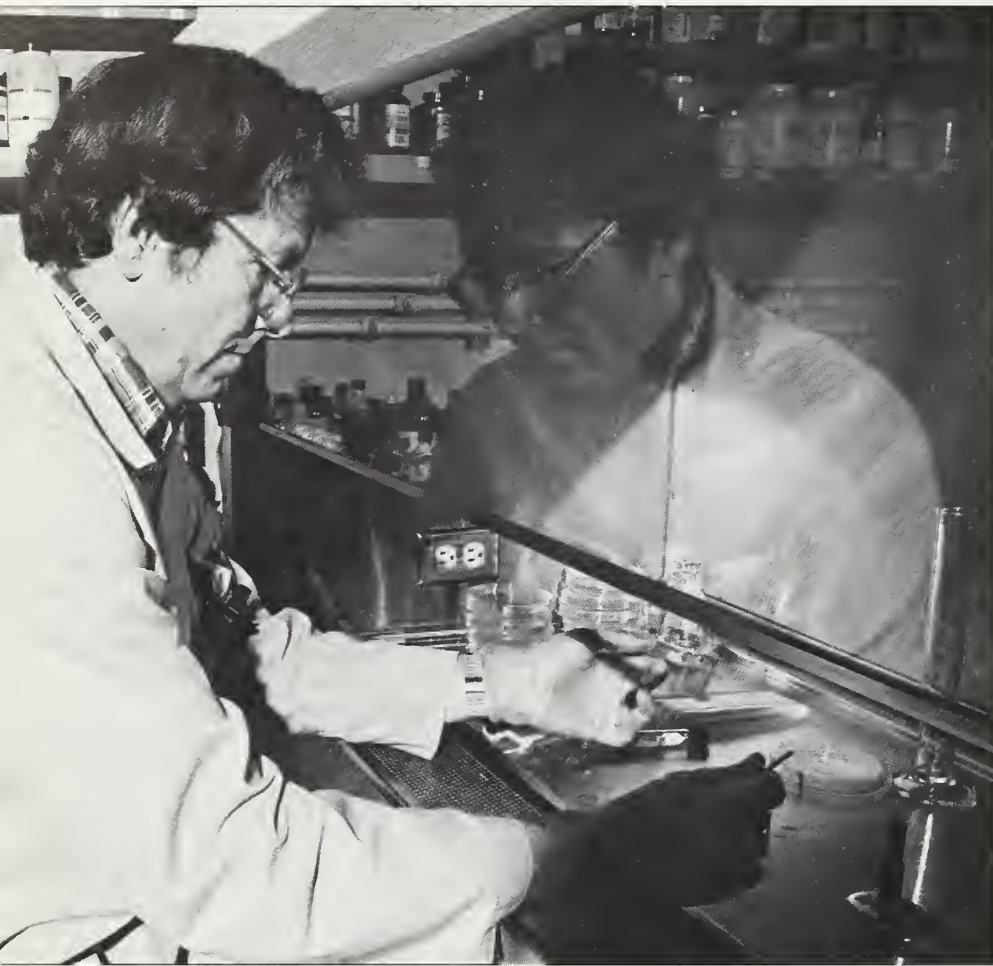
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Bean Gene Moved to Sunflower Cell



Above: Working under a safety cabinet, Dennis Sutton, ARS microbiologist, selects bacteria colonies that will transfer bean genes to sunflower cells (0681X596-2).

Right: University of Wisconsin lab technician Mary Ann Hansen measures density of recombinant bacterial extract before she purifies the protein-carrying DNA by centrifuge (0681X596-24).



A gene that directs production of a major seed protein has been moved from its native location in the French bean to the foreign environment of a sunflower cell.

"The genetic transfer is the first step in what may become an extremely versatile and useful model system for genetically engineering plants," says ARS biochemist John D. Kemp. Kemp and his research group worked with another group led by biochemist Timothy C. Hall, University of Wisconsin-Madison, to make the genetic transfer.

Highlighting the success of the research team was their observation that the transplanted gene is stable in its new environment and can produce small amounts of phaseolin-specific messenger RNA, the messenger that carries the genetic information from the gene to the protein synthesizing machinery of the cell.

The scientists transferred the gene for the seed protein, phaseolin, from the bean to fragments of a bacterium to sunflower tissue culture. The bacteria was *Agrobacterium tumefaciens*, a species that scientists often call nature's genetic engineer because it transfers a piece of its genetic material into plants that it infects.

The researchers inserted DNA containing the seed protein gene adjacent to the bacterial DNA that transfers bacterial genes into plants.

DNA or deoxyribonucleic acid is the genetic material that orchestrates the sustenance and duplication of living cells.

"The model genetic transfer system that we're continuing to study may open the way for creating genetic variations now unavailable because of sterility barriers between species and genera," says Kemp.

Scientists have long dreamed of using genetic engineering to develop new plants with increased nutritive value for human and animal nutrition, increased disease resistance, and nitrogen fixing capability.

In view of the research team's recent achievement, Kemp says some exciting challenges lie ahead.

"Before our genetic transfer becomes useful to agriculture other achieve-

ments must be made," says Kemp. "We might learn to mutate the bacteria so it still transfers DNA but not the genes that cause crown gall disease as *A. tumefaciens* normally does. Or we may learn to regenerate normal plants from cells that have been made abnormal with crown gall disease."

But there are many questions still to answer. Will the presence of the protein storage gene in the tissue culture increase appreciably the protein in the tissue culture? And if so, can the culture be made to produce sunflower plants with higher protein content than normal? No research team has developed the technology for propagating sunflower plants from tissue culture.

"What we do know is that we've made considerable progress in improving our understanding of a natural form of genetic engineering in which DNA is transferred from bacteria to cells of higher plants," says Kemp.

The researchers chose to work with the protein gene for a clear cut reason. Hall had studied the protein gene extensively, providing a framework of knowledge to help them succeed.

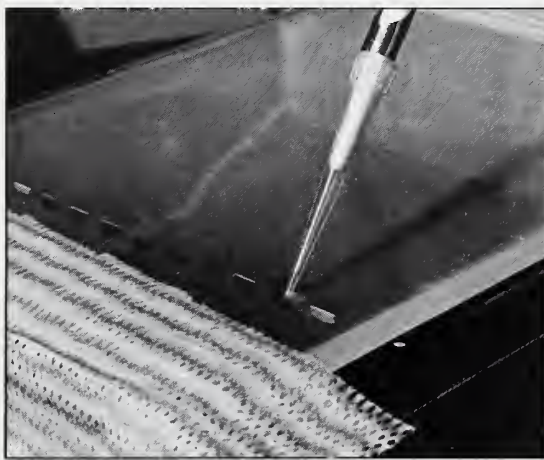
The protein that the gene causes bean plants to produce and store in their seed is a gamma globulin, a salt-soluble protein that animals easily digest.

Technical problems that the scientists solved may be followed by many more challenges before practical application is made in agriculture. But the possibilities may be substantial.

The research may provide many spinoffs to help lay the groundwork for agriculture in the 21st century. But it may be decades, for example, before researchers get nitrogen-fixing genes working in the genetic machinery of non-leguminous crop plants such as corn.

"Nitrogen fixation mechanisms in bacteria alone are controlled by many genes," Kemp says. "Disregarding complexities of DNA in the crop plant, the task of getting the genes produced and working together in the right combinations could become most difficult."

In the agricultural realm, recombinant DNA work may have its initial impact on crop plants that can be reproduced by some asexual means such as cuttings. Kemp is not sure whether sunflower plants can be engineered to produce



Left: After the DNA is separated from the bacterial extract and fragmented, fragments are separated by gel electrophoresis. In this purification process, "wells" in the gel are filled with DNA solution, and electrical charges flow through the gel to separate fragments according to size (0681X596-16).

Above: Kemp removes sunflower plant from growth chamber and examines it for recombinant tumors, in which gene localizes after *Agrobacterium* inoculation (0681X596-35).

seeds carrying the recombinant DNA.

"Perhaps incorporating a single foreign gene into the hereditary makeup of a plant will open the door to further advances in plant breeding by classical means," says Kemp.

Dr. Kemp's address is USDA-ARS Disease Resistance Laboratory, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706.—(By Ben Hardin, ARS, Peoria, Ill.)

Night Vision Goggles Aid Insect Trap Design



Above: Using a television camera and a monocular night vision device, Entomologist Wayne Shelton videotapes insect response to pink bollworm trap (1080X1354-5).

Night vision goggles and television cameras help ARS scientists study the nighttime activity of insect pests and improve the efficiency of traps for pink bollworm and tobacco budworm moths.

Traps are now being designed for night-flying insects by directly observing their response to the traps, according to ARS entomologists.

Traps are an important tool in integrated pest management and well suited for commercial use. They can monitor insect populations and determine the best time to apply control procedures. Live insects are also needed by research entomologists who use them to study the biology, ecology, and behavior of pests.

Pink bollworms and tobacco budworms are lured to traps baited with virgin females or synthetic insect sex lures (pheromones).

In nighttime studies of pink bollworm activity, Pete D. Lingren, ARS entomologist, Phoenix, Ariz., found that pink bollworm males, when entering a trap, tend to orient downward. Lingren pioneered the use of goggles for the nocturnal study of insect activity. A TV camera is used with a monocular night vision device to tape activity for later reference.

Two other ARS entomologists, Jimmy R. Raulston of Brownsville, Tex., and Alton Sparks of Tifton, Ga., using the same equipment, found that the tobacco budworm male, upon entering a trap, did the opposite; it oriented upward. Both the pink bollworm and the tobacco budworm, however, move from areas of low light levels to areas of greater light levels.

With the use of that and other information gathered from the study of night fliers, Lingren designed a pink bollworm trap made of plastic, and Raulston designed a wind-operated tobacco budworm trap made of galvanized steel and hardware cloth.

The Lingren trap consists of a lidded plastic funnel with holes cut into the sloping funnel sides. A clear plastic "jar" is glued to the spout to receive pink bollworm males. Synthetic pheromone is hung on the inside of the lid.

Males, attracted to the pheromone, enter through the holes in the side and

move downward from the relatively dark interior to the area of greater light intensity—the clear plastic jar—and are captured. The Lingren trap captured 3 to 15 times more pink bollworms than traps of existing designs.

A wind vane on the Raulston trap keeps it headed into the wind with the pheromone “plume” trailing downwind. The tobacco budworm males pick up the “scent,” find their way to the trap, enter the orifice, and move upward into the live box where they are captured. When first designed, the trap had a complete bottom. Night observations showed, however, that the budworms, attracted in this test by virgin females, refused in many instances to enter the trap. That observation, coupled with observations that the angle of approach was from the downward side of the trap, prompted Raulston to remove nearly half of the bottom.

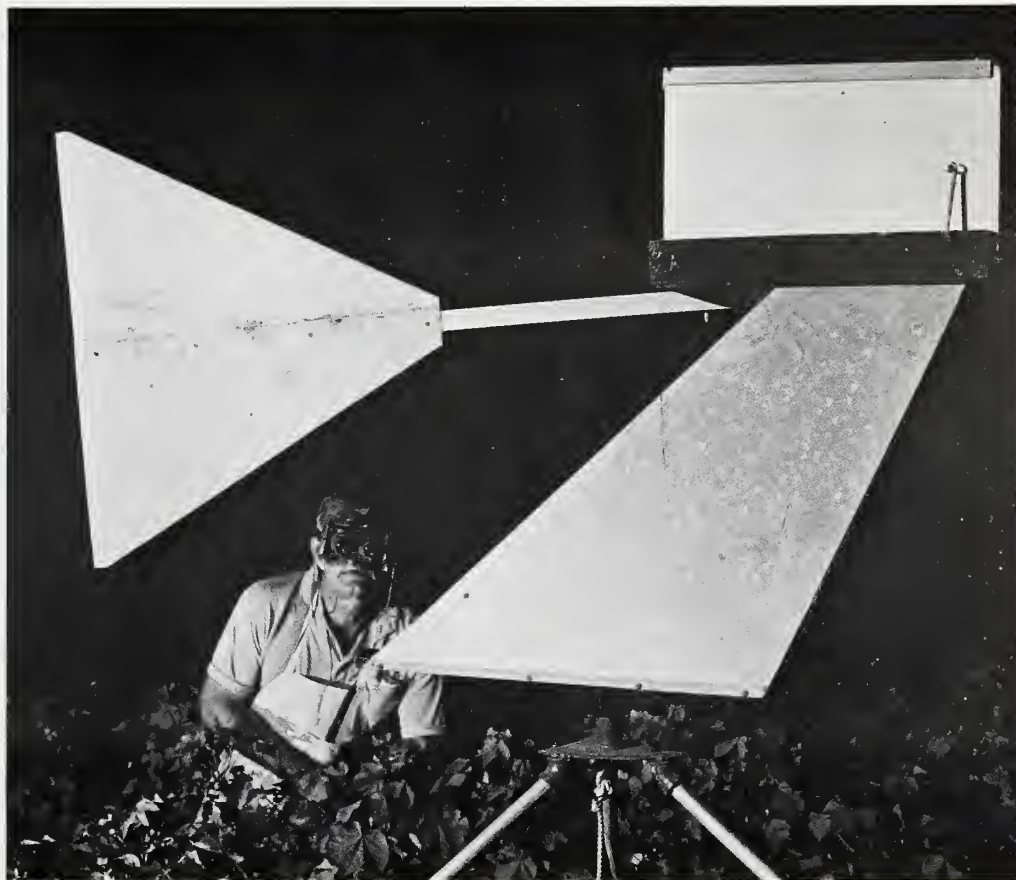
That modification increased the efficiency of the trap more than four times. The trap as first designed captured 182 moths while the modified trap captured 855.

“The plastic trap appears to have considerable potential for commercial use as a standardized trap for monitoring and evaluating pink bollworm populations in the field,” says Lingren.

He added that further studies of other nocturnal insect pests using the techniques developed in the research studies should result in the design of a standardized pheromone trap for several other insects.

“Use of the new trap, or other traps like the Raulston trap, developed as a result of these tests should allow the cotton producer to more accurately time and assess control procedures for pink bollworms and tobacco budworms resulting in less cost than now,” says Lingren.

Dr. Lingren is located at the ARS Western Cotton Research Laboratory, Phoenix, Ariz.; Dr. Raulston is located at the ARS Cotton Insects Research Laboratory, Brownsville, Tex.; and Dr. Sparks is located at the ARS Cotton Insects Research Laboratory, Tifton, GA.—(By Paul Dean, ARS, Oakland, Calif.)



Terrence Myers, biological technician, counts pink boll worm catch. The count helps scientists make more accurate estimates of insect population dynamics (1080X1353-16).



Top: By intensifying natural light up to 100,000 times, night vision goggles enable Peter Lingren, ARS entomologist, to monitor nighttime insect response to tobacco budworm trap. Pheromone lures budworm males into trap, and they are captured in the live box, top (1080X1354-26).

Above: Lingren inspects pink bollworm trap. Tricked by pheromone bait seen through hole in funnel, bollworm males enter funnel and are trapped inside the clear plastic container (1080X1352-32).

Micropropagation Speeds Up New Peach Varieties

A new plant propagation technique, micropropagation, could result in new varieties of peach trees many times faster than currently possible, says Freddi Hammerschlag, ARS plant physiologist, Beltsville, Md.

It now takes 6 or more years of testing before new varieties can be released to the public. However, successful micropropagation of peach shoots should cut testing time in half by providing large numbers of identical plants in a relatively short period of time.

Micropropagation of peaches will also facilitate high-density orchard management in which 500 to 700 small trees are planted on each acre instead of the usual low-density of 150 to 180 trees, says Hammerschlag. Although this high-density management has been shown to be more productive and potentially less costly per acre, peach trees produced by the older method, bud grafting, have been too expensive and too large to be used in this new system. However, rooted shoots from micropropagation are small and inexpensive enough for high-density orchard management, she says.

Traditionally, peaches have been propagated by bud grafting, because peach seeds do not reproduce all the characteristics of parent trees, and peach cuttings from most varieties are very difficult to root. In the old grafting technique, dormant buds are inserted into the stem of a few types of peaches produced from seed. This procedure is costly, time consuming, and produces only one plant per bud.

With Hammerschlag's micropropagation system, a tiny shoot from budwood can produce 5 to 10 shoots every 6 weeks. Then each new shoot can be used to produce many more shoots. The shoots are then encouraged to root and are put into soil for shipment to growers. This procedure can be initiated at any time of year and carried out in a limited space.

In the past 5 years, plant tissue culture facilities have sprung up all over the world. In Europe, for instance, England, Belgium, and Italy are heavily involved in micropropagation of woody species.



During this period, techniques have been worked out for propagation of at least 50 woody plant species. Research is being supported by both public and private funds.

The major commercial efforts in micropropagation in the United States are in California and Florida where some facilities sell plants in tubes as novelty items. At the Beltsville Agricultural Research Center, micropropagation of apple, blackberry, blueberry, strawberry, and *Prunus* species is being done.

Working in the Cell Culture and Nitrogen Fixation Laboratory, Hammerschlag micropropagates peach plants from axillary shoots because they produce plants identical to the parent. This procedure is divided into three stages. First, shoots are forced to grow from cold-treated dormant budwood, or shoots are removed from actively growing plants and then reduced in size. The

tiny shoot tips are established on a sterilized culture medium with plant hormones or synthetic plant growth regulators that induce shoot growth. Shoot tips are then grown under controlled temperature and light conditions for 3 to 6 weeks.

Second, shoots are transferred to test tubes for 1 week, and then moved to glass jars where multiplication takes place.

Finally they are transferred to a rooting medium with a growth regulator that induces roots to grow. Rooted plants are grown in pots until they are ready for testing in the field.

The development of different plant organs in tissue culture is controlled by groups of hormones, known as auxins, cytokinins, and gibberellins. Balance between auxins and cytokinins determines type of growth and organ formation. When cytokinin levels are higher, shoots are produced. If auxin levels are higher, roots are produced.

Hammerschlag has worked with many popular peach varieties, including Dixi-



red, Jerseyqueen, Redskin, and Sun-high. She is now testing different combinations of temperature, mineral salts, auxins, and gibberellic acid to find optimum conditions for rooting peach shoots. Thus far, her studies indicate that the optimum temperature for tissue culture propagation is 70° to 75°F and that peach plants grow best on a liquid culture medium.

In addition, Hammerschlag is cooperating with Ralph Scorza, ARS plant geneticist, Appalachian Fruit Research Station in West Virginia to compare yield, vigor, tree size, and genetic stability of micropropagated fruit trees to that of trees propagated by grafting.

Dr. Freddi Hammerschlag is located at the ARS Cell Culture and Nitrogen Fixation Laboratory, Rm. 130, Bldg, 011-A; BARC-West, Beltsville, MD 20705.—(By Ellen Mika and Marguerite Benedict, ARS, Beltsville, Md.)

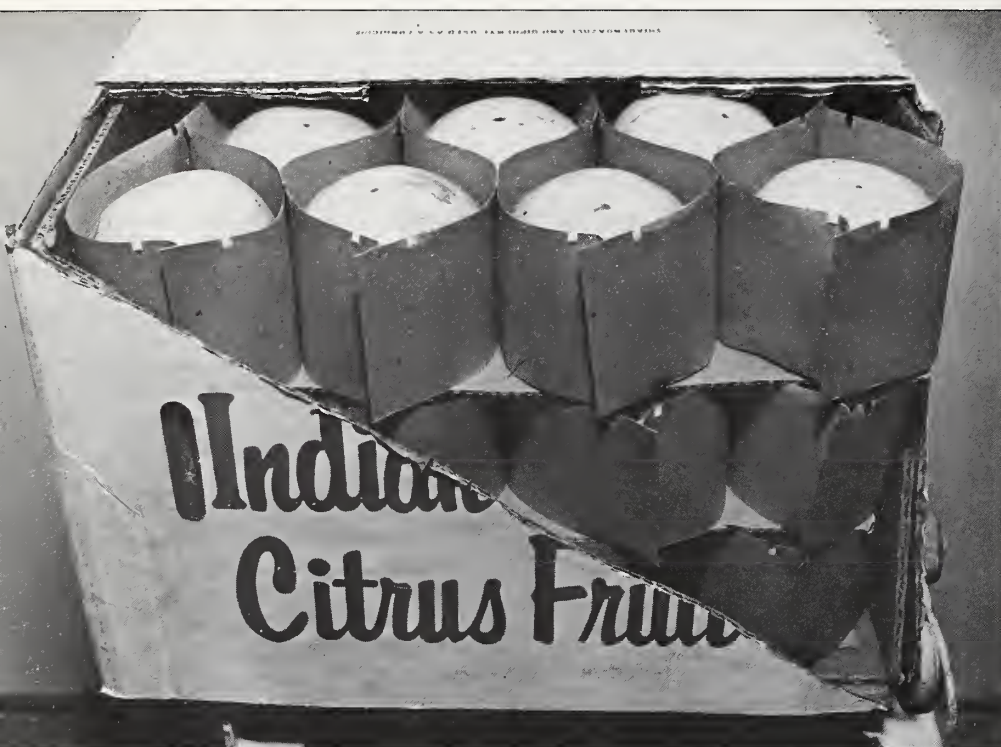


Above: Freddi Hammerschlag, ARS plant physiologist, snips peach budwood for micropropagation (0281W140-11a).

Left: After shoots are forced from budwood, shoot tips are established on sterile culture media. The filter wick in this vial feeds nutrients and plant growth regulators to a Jerseyqueen peach shoot (0281W139-21a).

Opposite: Growth regulator levels determine how peach shoots will develop during micropropagation. Hammerschlag observes shoots that regulators have induced to multiply (0281W139-3).

Shipping Grapefruit — A Success Story



Above: This new cell-pack honeycomb box has reduced dramatically damage to grapefruit during test shipments to Japan. Thin chipboard honeycombs and fiberboard layer dividers ease pressure on fruit (0681X595-15).

Most people tend to “eat with their eyes,” says agricultural marketing specialist Philip W. Hale. They avoid fruit that has become deformed during packaging and shipping.

Working at the U.S. Horticultural Research Laboratory, Orlando, Fla., Hale has developed a new honeycomb, cell-pack box for shipping grapefruit and ensuring their near-perfect condition on arrival in Japan.

Fresh fruit exported from the United States accounts for 72.2 percent of the estimated 420,854 metric tons of fresh oranges, limes, grapefruit, pineapples, melons, and other fresh fruit imported by Japan. Since the time required for storage and transport of fresh grapefruit—a highly priced delicacy in Japan—averages 38 days, the fruit must be packaged and handled with care in order to arrive in top condition.

In 1972, one year after Japan liberalized its import restrictions on

citrus fruit, researchers at the U.S. Horticultural Research Laboratory began studying packing materials that would provide increased protection for grapefruit during transit from packinghouse to Tokyo.

Hale, John J. Smoot, plant pathologist, and Thurman T. Hatton, Jr. horticulturist, evaluated 15 types of shipping containers. Results showed that regardless of container type, the amount of seriously deformed fruit ranged from 33 to 60 percent. Seriously deformed fruit is defined as that on which the flattened or indented surface area equals 2 or more inches in diameter.

In controlled laboratory tests, Hale found that the higher the fruit bulged over the top when packed, the more serious the deformation.

In 1977, Smoot and Hale initiated commercial testing of boxes one-half inch deeper than standard boxes. This extra depth enabled fruit to be packed without bulge and reduced the amount of serious deformation to 12.1 percent—a dramatic reduction from the former high of 60 percent.

All shipping tests showed that deformed fruit was more prevalent in the bottom layer of the boxes, and observations of commercial shipments indicated that large fruit suffered more deformation than smaller fruit.

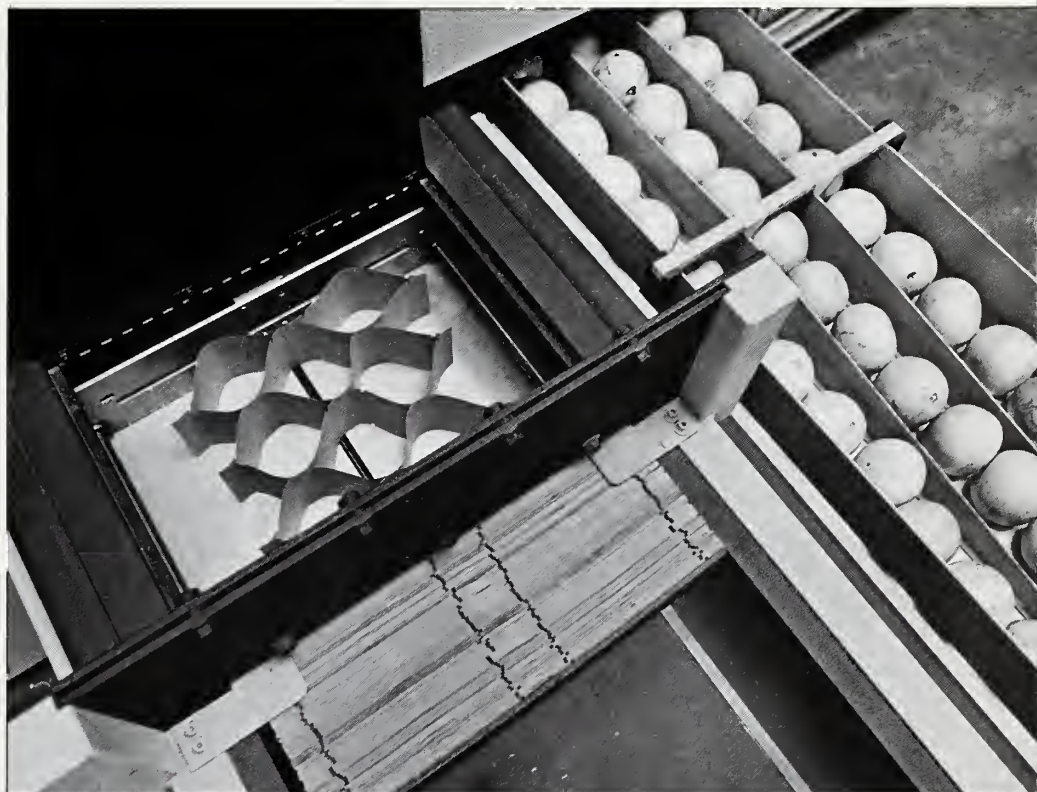
“Large, well-shaped fruit brings a premium price in Japan—at one time as much as \$2 per grapefruit,” says Hale. “And shippers and receivers have long wanted a package that would deliver grapefruit in optimum condition. A recent study was limited to testing cell-pack boxes for large fruit only, those that had an average diameter of 4¼ inches. Three export tests were made from Tampa, Fla., to Tokyo, Japan.”

The cell-pack box is a full-telescope, single-wall fiberboard box with inside dimensions of 16-5/8 by 11-1/2 by 12 inches. Each side panel of the box has two ventilation slots and each end panel has one ventilation slot.

The shipping container was packed with three layers of grapefruit, blossom end up, and 11 fruits per layer—one more fruit per container than in the standard 4/5-bushel, place-pack box.

A single-wall honeycomb partition, 0.017-inch-thick chipboard material that expanded and formed individual cells 3.75 inches high for each grapefruit, was used for each layer. Single-wall fiberboard dividers were placed below and above the middle layer of fruit. The support provided by the honeycomb partitions, in combination with the fiberboard layer dividers, prevented overhead pressure on each layer of fruit. Two biphenyl pads were placed in the box to prevent green mold, one between each layer of fruit.

After packing, all the boxes, along with three standard 4/5-bushel boxes, were unitized with commercial grapefruit on wooden pallets for shipment. For test purposes, these boxes were placed in the bottom layers of unitized stacks of boxes seven layers high because fruit and boxes on the bottom layer take the most punishment.



As required by the Japanese government, the grapefruit were fumigated with ethylene dibromide at the Florida Division of Plant Industry Station near Bartow, Fla., before they were loaded into the refrigerated holds of ships at Tampa.

Examination of the fruit received in Tokyo proved the success of the new packing technique. All degrees of deformation were less for the grapefruit shipped in the cell-pack containers than for those shipped in standard boxes.

The cost of the cell-pack container, including packaging materials, labor, and transportation, is 3 cents more per fruit than the standard box. However, Japanese importers believe the amount of money received by retailers for premium fruit in near-perfect shape should offset any difference in cost.

The honeycomb packages are now being evaluated in commercial-sized lots, says Hale.

Mr. Hale, Dr. Smoot, and Dr. Hatton, Jr., are located at the U.S. Horticultural Research Laboratory, 2120 Camden Road, Orlando, FL 32803. —(By Peggy L. Goodin, ARS, New Orleans, La.)



Above left: Before an automatic packing machine was designed for the new boxes he developed, Philip Hale, agricultural marketing specialist, had to pack them manually. Here Hale packs grapefruit for the initial test shipment to Japan (0681X595-12).

Above: The automatic packing machine began operating in time for the second season of test shipments. Grapefruit line up in the feeding shoot for placement into honeycombs (0581X471-16).



Left: Rubber cups lift grapefruit in pre-packing position and will lower them into expanded honeycomb, which will then be lowered into box (0581X472-6a).



Lower left: Test boxes, unitized with commercial boxes on wooden pallets, await loading onto the *Sunbelt Dixie* for their voyage to Tokyo (0381X476-33).

Natural Products Repel Cucumber Beetle



Keeping insects from eating crops is hard enough, but to keep them from even taking a nibble is the real test. That's the goal ARS entomologist David K. Reed has set for himself at USDA's Fruit and Vegetable Insects Research Laboratory, Vincennes, Ind.

"We are trying to develop natural products for use as nontoxic, safe, and biodegradable alternatives to chemical pesticides. We hope to prevent the insects from feeding on cucurbit crops by influencing their behavior," says Reed.

The striped cucumber beetle is not satisfied with just eating the stems, leaves, blossoms, and fruit of cucumbers, squash, muskmelons, and zucchini plants; it also spreads bacterial wilt.

The scientist is testing two extracts of the seeds of neem, a tree common to India, as an antifeedant. Both extracts, azadirachtin and salannin, were highly active against striped beetle feeding, he says.

If neem extracts could be used to inhibit feeding by the striped cucumber beetles, particularly on muskmelon, feeding and disease losses, sometimes ranging as high as 25 percent in the north central states, could be reduced. The destructive feeding damage is significant, but the ability of the insect to transmit bacterial wilt is even more damaging. In southern Indiana alone, bacterial wilt causes 5 to 15 percent loss of the muskmelon crop every year, according to Reed, at a cost of between \$400,000 and \$1 million.

"Natural materials such as neem extracts offer several advantages," Reed says. "Chemical insecticides may harm bees and other pollinators, and during dry weather repeated applications may build up to the point of being toxic to crop plants."

Developing muskmelons with resistance to wilt, another project underway at the laboratory by coworker Gary L. Reed, ARS entomologist, and

using them in combination with an effective antifeedant, might provide protection for the plants without need for chemical pesticides. Such a system would be especially valuable early in the season because delaying the establishment and spread of wilt until later greatly alleviates the effect on yields.

Reed tested the neem extracts by cutting pieces from cantaloup leaves, dipping them in the extract solution, and placing them in a dish with untreated leaf pieces. He then added five beetles that had not eaten for 24 hours. Some concentrations protected the leaves from beetle feeding for 22 hours, he says.

In another test, Reed sprayed muskmelon seedlings and put them in a cage with 24 striped beetles. The 0.1 percent azadirachtin application provided good protection for 3 days and some protection even 10 days after treatment.

Because bacterial wilt is often transmitted to seedlings in beds or cold frames, diseased plants are sometimes inadvertently transplanted into the field where further beetle feeding spreads the disease to healthy plants. To see if neem extract would protect seedlings, Reed sprayed trays of seedlings with the extract and added 300 striped cucumber beetles which had fed on wilt-infected plants for 14 days. After 2 days, he removed the beetles and checked the plants for feeding damage; then after 24 days, he checked for symptoms of bacterial wilt.

Azadirachtin applications of 0.25 percent concentration protected the plants from disease symptoms though they did show some minor feeding damage, Reed says.

The scientist believes results might be improved by increasing concentrations, using subsurface irrigation to avoid washing the antifeedant off the plant, using an additive to help the material stick on the leaves, or applying the material through the plant roots with a trickle irrigation system.

Dr. David K. Reed and Dr. Gary L. Reed are located at the USDA Fruit and Vegetable Insects Research Laboratory, P.O. Box 944, Vincennes, IN 47591.—(By Ray Pierce, ARS, Peoria, Ill.)

Wheat Research Pays Big Dividends

Investments in wheat breeding research at Purdue University have paid off by increasing soft red winter wheat production about 1 billion bushels since 1946.

The farm value of this additional wheat—enough to make 54 billion 1 pound boxes of crackers—amounts to more than \$1.8 billion. The retail value would exceed \$50 billion.

ARS agronomist John J. Roberts at Purdue, West Lafayette, Ind., says the increased yield calculations are based on comparisons of yields of an old popular variety, Trumbull, with yields of 19 newer varieties that ARS and Purdue University scientists jointly developed and grew alongside Trumbull.

"In each new variety we have made improvements in pest resistance and other agronomic qualities," says Fred L. Patterson, Purdue agronomist.

Wheat varieties developed since 1946 by the Indiana research team have produced about 4.3 billion bushels. These varieties now grow on more than 75 percent of the acreage planted in soft red wheat in the eastern United States. Some of the more popular varieties over the years have been Knox, Monon, Redcoat, Arthur, Arthur 71, Abe, Oasis, and Sullivan.

What can the wheat researchers do for an encore?

"One thing we're trying to do is to develop new varieties with resistance to barley yellow dwarf disease," says Roberts. This disease afflicts many types of cereal crops all over the world.

"Although the disease often produces hardly noticeable symptoms in wheat, I think it probably always reduces wheat yields in Indiana by 5 percent," says Roberts.

The research team has introduced into a breeding line of wheat a chromosome from tall wheatgrass that imparts resistance to barley yellow dwarf disease. This was accomplished by use of genetic techniques worked out by cytogeneticist Ernest R. Sears, Columbia, Mo., now retired from ARS.

Another disease that Roberts and his colleagues are challenging through breeding work is loose smut. Roberts



envisioning future varieties of wheat that might resist loose smut infections by keeping their blossoms tightly closed. A hope for this development is spurred by the success that Purdue and ARS scientists have already had in breeding cereal grains with hairy leaves that repel insects.

Dr. John J. Roberts is located in Rm. 2-310, Lilly Hall of Life Sciences, Purdue University, West Lafayette, IN 47907.—
(By Ben Hardin, ARS, Peoria, Ill.)

Wheat research—growing more and growing stronger. (Photo courtesy of Grant Heilman.)

Opposite: Two neem seed extracts are being tested as an antifeedant to help make this striped cucumber beetle avoid his favorite foods. (Photo courtesy of Grant Heilman.)

Advances in Vitamin D Metabolism Research

ARS physiologist Ronald L. Horst is one of the few scientists who can accurately measure most of the plant and animal forms of vitamin D and their known biological end products in animals and humans.

In 3 years, Horst's analytical techniques have replaced estimating vitamin D concentrations in living systems and have had a major impact on animal nutrition, veterinary medicine, and human nutrition.

In animal nutrition, methods of supplying supplemental vitamin D have been reassessed and improved.

In veterinary medicine, studies with Horst's techniques identified previously unsuspected susceptibility to rickets in confinement-raised newborn pigs and calves. Also, more effective vitamin D therapy for milk fever and grass tetany, diseases of mineral metabolism in cattle, is now possible.

In human medicine, collaborative studies are providing new insight into the treatment of diabetes, malignancies, hyperparathyroidism, and bone diseases.

Basic and applied studies of mineral metabolism using Horst's techniques are answering important questions about mineral metabolism.

At the basic research level, these have included isolation and identification of previously undescribed biological end

products (metabolites) of vitamin D, definition of vitamin D metabolic pathways in pigs and cattle, and evaluation of vitamin D's role in spontaneous diseases of mineral metabolism in animals and humans.

In the past, Horst explains, the crude, laborious technique for estimating vitamin D concentrations in living systems limited the usefulness of vitamin D analysis in clinical investigations. His highly sensitive techniques can accurately measure vitamin D₂, the plant form, and vitamin D₃, the animal form, and their 11 metabolites in blood and milk of animals and man.

Horst and E. Travis Littledike, veterinary medical officer, found that swine and cattle preferentially utilize the animal form of vitamin D, but sheep use both forms equally well. Only chickens and new world monkeys were previously thought to discriminate between the forms. Current practices, now being revised, for adding supplemental vitamin D in plant form to swine and cattle rations assumed equal use of both forms.

The researchers, cooperating with Iowa State University's Nutritional Physiology Laboratory, also found that giving vitamin D to ruminants orally, a current practice, is ineffective.

Borderline rickets may exist in newborn pigs raised in confinement, Horst and associates found, even though the sows had been supplied with vitamin D in accordance with National Research Council recommendations. The deficiency can be corrected by exposing the pigs to ultraviolet light or administering vitamin D to the sow.

Similarly, confinement-raised newborn calves receiving alfalfa silage are very susceptible to rickets, the researchers found. Preparation of alfalfa silage produces a feed virtually devoid of vitamin D activity, they say.

Insight into timing and dosage levels for vitamin D from research by Littledike and Horst now sets the stage for devising safe, effective measures for preventing or controlling milk fever and grass tetany.

Administering vitamin D to prevent or control these diseases has been relatively ineffective or has caused toxicity. Direct costs of losses from milk fever and grass tetany are estimated at more than \$30 million annually in this country. Indirect costs may be many times this amount.

In collaborative studies with the Yale University Medical School, New Haven, Conn., precise measuring of vitamin D in humans has demonstrated:

- Diabetic humans have low blood levels of the major biologically active form of vitamin D, possibly explaining the frequent development of bone disease in diabetics.

- Patients with malignancies likewise have low levels of this vitamin D metabolite in the blood. An unknown factor acting on bone may be responsible for the high blood calcium frequently complicating malignancies.

- Patients with primary hyperparathyroidism can be separated into two groups, requiring different therapy, by the concentrations of biologically active metabolites in their blood.

- The biologically active form of vitamin D increases during adult-onset osteodystrophy. This finding may alter the therapy given these patients.

Patients receiving intravenous feeding at the Wadsworth Veterans Administration Hospital, Los Angeles, Calif., proved unable to synthesize biologically active vitamin D, in another collaborative study. This observation explains the development of bone disease in these patients and has important implications in their treatment.

Dr. Horst and Dr. Littledike are located at the National Animal Disease Center, P.O. Box 70, Ames, IA 50010.—
(By Walter Martin, ARS, Peoria, Ill.)

Agrisearch Notes

Monarch — A New Cicer Milkvetch.

ARS scientists have developed a new variety of cicer milkvetch, called Monarch, that has up to twice the seedling emergence of Lutana, currently the most popular cicer milkvetch.

High seedling emergence is important for plant establishment on irrigated and dry pastures and on strip-mined areas. The new legume also has a forage yield that equals or exceeds that of Lutana. Nutrient and feeding values of monarch are equal to alfalfa, but unlike alfalfa, cicer doesn't cause bloat in livestock.

Cicer milkvetch is one of the better legumes for revegetation of strip-mined areas at the higher elevations—above 6,000 feet. There is some reluctance to seed alfalfa in these areas because of potential bloat problems.

Monarch, also, is adapted to the northern and central Great Plains where annual precipitation exceeds 16 inches. In addition, it performs well on irrigated pastures.

Like alfalfa, cicer milkvetch, in association with the proper bacteria, fixes its own supply of nitrogen on roots so that nitrogen fertilizer is not required for its growth. This feature is particularly important because mine spoils are very deficient in nitrogen.

Like other cicer milkvetch varieties, Monarch can be seeded alone or in combination with cool season grasses such as brome grass, according to Charley Townsend, an ARS plant geneticist at Fort Collins, Colo.

Cicer milkvetch, native to southeastern Europe and southwestern Asia, is relatively new to the United States, having been introduced here in the 1920's. Cicer is well-suited for grazing because most of its regrowth comes from axillary buds near the base of the stems, and its rhizomes permit rapid spread under favorable conditions. Because of these attributes, cicer milkvetch has the potential for increasing the productivity of many acres of rangelands and dryland pastures.

Foundation seed of Monarch was made available to seed growers in 1980. Commercial quantities of seed should be available to farmers and ranchers this fall.

ARS developed Monarch and released it in cooperation with the Colorado State University (CSU) Agricultural Experiment Station.

Dr. Townsend is located at the Crops Research Laboratory, CSU, Fort Collins, CO 80523.—(By Dennis Senft, ARS, Oakland, Calif.)

Integrated Control of Lone Star Ticks.

Integrated control of lone star ticks on cattle is the goal of a new research facility at Poteau, Okla.

Gary A. Mount, ARS entomologist, says that the new facility plans to test the integrated use of vegetative management, animal management, and acaricides (chemical control agents) to control lone star ticks.

Scientists will also study the ecology of the lone star tick to determine the times of the year and the environmental conditions that would enable the most effective use of minimum quantities of acaricides.

Mount says that the abundance of lone star ticks in the south-central United States, especially in the Ozark-Ouachita Highlands, causes great economic loss to agricultural and recreational industries.

Homeowners and private landowners suffer as well, since the bloodsucking pest will feed on pets, wildlife, and human beings as well as livestock.

Scientists will focus their studies on recreational areas where results from combined vegetative management and pesticides can best be measured. They hope to develop a system to control this costly and troublesome pest.

Mr. Mount is located at the Lone Star Tick Research Laboratory, P.O. Box 588, Poteau, OK. The Lone Star Tick Research Laboratory is a sublaboratory of the U.S. Livestock Insects Laboratory in Kerrville, TX.—(By Bennett Carriere, ARS, New Orleans, La.)

New Potato Variety.

Russet Burbank faces another challenge to its current position as the United States' top potato. ARS researchers have developed a potato that outyields Russet Burbank, is higher in vitamin C, and is more resistant to malformation.

The new variety, Lemhi Russet, was developed by ARS geneticist Joseph J. Pavék and plant pathologist Dennis L. Corsini, Aberdeen, Idaho. They also developed the popular Butte potato released in 1977.

The new variety produces attractive, russet-skinned tubers that are uniformly oblong in shape and blockier and larger than Russet Burbank potatoes. Test yields have been 34 percent higher in U.S. Number 1 tubers than for Russet Burbank. Lemhi Russet is also 6 percent higher in solids, 50 percent lower in sugars that build up in storage, and 22 percent higher in vitamin C.

Russet Burbank growers who've had problems with net necrosis should welcome Lemhi Russet which has excellent resistance to both net necrosis and common scab, and is equal to Russet Burbank in resistance to early blight and Verticillium wilt.

Lemhi Russet possesses excellent internal quality. Its mealy texture makes it an outstanding potato for baking and processing into french fries though it is poor for boiling. It is more susceptible to bruising and hollow heart than Russet Burbank.

Dr. Pavék and Dr. Corsini are located at the University of Idaho Research and Extension Center, Aberdeen, ID 83210.—(By Lynn Yarris, ARS, Oakland, Calif.)

States with Good Wind Energy Potential.

States best suited to the economical use of wind energy for irrigation pumping are Texas, Oklahoma, Kansas, Nebraska, Colorado, and New Mexico,

(Continued on page 16.)

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says ARS agricultural engineer Nolan Clark.

"Those six states," says Clark, "use 63 percent of the total energy consumed to pump water in the United States. We've been doing studies to match wind power with irrigation pumps and the best correlation is found in those good wind energy states."

Rising energy costs are causing farmers to consider alternatives to fossil fuels for pumping irrigation water. One of those alternatives is wind energy. However, for wind energy use to be practical economically, it must be used year round. When wind is not used for pumping irrigation water, its energy must be used in other ways, perhaps as electricity for home use and for other farm uses such as drying grain, or even for sale to power companies.

The machine used for "harvesting" wind is the wind turbine, which differs significantly from the old-fashioned windmill. Turbines have only two or three blades and operate on the principle of lift, as airplanes do, rather than the simple pushing power used by traditional windmills.

Recently, irrigated land using pumped water in the United States reached 44 million acres requiring some 473,000 pumps. Clark says the potential for the conservation of expensive fossil fuels and for economic benefits through harvesting the wind is very great.

Dr. Clark is located at the Great Plains

Research Center, Bushland, TX 79012.—(By Bennett Carriere, ARS, New Orleans, La.)

Heifers Test Fescue Quality.

Beef heifers grazing on small pastures of Kenhy or Missouri 96 tall fescue grass varieties generally gained about 50 percent more weight than heifers grazing on an older and more common variety, Kentucky 31.

ARS scientists at Columbia, Mo., who made this observation from spring, summer, and fall tests, kept the grazing pressures in the pastures as uniform as possible. They tried to keep daily herbage availability at about 2.5 percent of body weight throughout the experiments which ranged from 35 to 68 days.

"We were testing the quality of the grass rather than its yield," says agronomist Arthur G. Matches. The research was aimed at developing improved techniques for screening breeding lines of grasses.

Matches says forage quality measurements in the laboratory may someday be as reliable for predicting animal performance as current grazing experiments. Until then, he and his colleagues at the University of Missouri-Columbia plan to give the animals a role in deciding what is best for them. However, additional research may be needed to determine the best herbage allowance for measuring forage quality differences, Matches says.

Dr. Matches is located at the University of Missouri, Rm. 207 Waters Hall, Columbia MO 65211.—(By Ben Hardin, ARS, Peoria, Ill.)

Porcine Parvovirus Immunity.

The effectiveness of vaccinating gilts against reproductive failure caused by porcine parvovirus depends on the time of vaccination as much as on the quality of the vaccine.

Pigs get antibodies during the first few days of their life by ingesting colostrum and milk from mother sows that are immune. These passively acquired antibodies may persist for several months and interfere with the development of active immunity after the pigs are vaccinated.

Prem S. Paul and William L. Mengeling, ARS veterinary medical officers, found that passively acquired antibodies for porcine parvovirus lasted as long as 6½ months when large amounts were absorbed from colostrum.

Since most gilts are initially bred when they are 7 to 9 months of age, the scientists recommend that the vaccine not be administered until about 2 weeks before breeding, to allow time for active immunity to develop before conception.

Dr. Paul and Dr. Mengeling are located at the National Animal Disease Center, P.O. Box 70, Ames, IA 50010.—(By Walter Martin, ARS, Peoria, Ill.)